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PHYSIOLOGICAL SENESCENCE IN HYDROMEDUSÆ.

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The following observations on senescence in the hydromedusæ were made at the Puget Sound Marine Station, Friday Harbor, Wash., during the summer of 1917. I wish to acknowledge my obligation to the director of the laboratory, Dr. Frye, for the use of a room and the privileges of the station, and for his kindness in providing special glassware and other equipment. To other members of the staff I am also deeply indebted for many courtesies.

The great abundance, variety and large size of hydromedusæ at Friday Harbor makes this an extremely favorable locality for work on these forms. During the summer of 1917 five species of hydromedusæ, *Phialidium gregarium*, *Aequorea cærulescens*, *Sarsia rosaria*, *Mitrocoma discoidea* and *Stomatoca atra* could readily be obtained in considerable numbers, often by the hundred, from the laboratory float, and five other species of other genera were more or less abundant, besides two species of siphonophores. The first four species named above furnished the material for the observations and experiments on ageing, and the results are essentially the same in all.

The work was done entirely on free-swimming medusæ and is essentially a comparison of younger and older individuals, no attempt having been made to obtain the medusa-bud stages before detachment from the hydroid.¹ In the selection of individuals of different age for experiment, size of the animal and degree of development of the gonads were used as superficial criteria of age. As might be expected, these two criteria are in general in close agreement, for since the range in variation in size of the medusæ-bud at a given stage of development is not very great, a larger medusa must in general have undergone a greater amount of growth than a smaller, and may therefore be

¹ In *Pennanria* a progressive change in physiological condition has been observed from earlier to later stages of medusa-buds (Child, '15, pp. 150-152).

expected, if age differences exist, to be physiologically older. As has been shown for *Planaria*, and other simple animals (Child, '15, Chapter IV.—VII.) physiological age is not necessarily a function of the length of time which the individual has lived, for nutrition, agamic reproduction and various other factors play a rôle in determining the physiological age at any given time. Size is therefore a far more adequate criterion of age in such forms than time. Moreover, since the development of the gonads in medusæ progresses from a relatively early stage to sexual maturity the condition of the gonads also serves as a measure of the stage in its life history which the animal has reached.

The chief method employed was that of determining the susceptibility of younger and older animals to various agents. The general relation between susceptibility and age has been discussed elsewhere (Child, '14, '15) and need only be briefly restated. In general, in concentrations or intensities sufficiently high to kill without permitting acclimation, the susceptibility decreases with advancing age. In low concentrations, which permit some degree of acclimation, the susceptibility increases with advancing age because the young individual possesses a greater capacity for acclimation than the older, therefore becomes more rapidly and more completely acclimated and so dies later than the older, or lives indefinitely, while the older dies sooner or later.

It has been shown that a relation exists between susceptibility and metabolic activity, more particularly the oxidative or energy-freeing reactions. In general, susceptibility to the higher concentrations or intensities varies directly, and susceptibility to the lower concentrations varies inversely as the rate of these reactions, because acclimation occurs more rapidly and more completely where metabolic activity is greater. This relation between susceptibility and metabolic condition has been shown to hold good not only for different individuals, *e. g.*, young and old (Child, '15) but for different regions of the body of a single individual (Child, '16a).

It does not follow, however, from the existence of a relation between susceptibility and rate of metabolism or of certain metabolic reactions, that all agents for which such a relation

exists act directly upon the chemical reactions or that they all act in the same way. Much remains to be learned concerning the nature of the action of external agents on living protoplasm, but the fact is already clear that protoplasmic structure, aggregate condition, permeability, surface tension, etc., are associated with metabolic condition. Susceptibility to a given agent serves at best merely as an indicator of protoplasmic and metabolic condition and tells us nothing concerning the way in which the agent acts. As our knowledge of susceptibility to different agents and conditions increases, we find that under certain conditions different agents give us different results, and such differences in susceptibility relations often serve to throw additional light on the physiological condition of the protoplasm or the action of the agent, or both. The general relations stated above hold for a wide range of agents and conditions, and it is primarily with these general relations that we are concerned in the present paper.

AGE-DIFFERENCES IN BEHAVIOR.

A very conspicuous difference between young and old individuals is the much higher rate of pulsation in the young animal and its progressive decrease with advancing age. Table I. gives characteristic data on this point.

TABLE I.

Species.	Diameter of Body in Mm.	Pulsations in 20 Sec. at 14° C.
<i>Æquorea cærulescens</i>	7	40
	20	32
	50	20
	18	28
<i>Mitrocoma discoidea</i>	60	16
	5	40
<i>Sarsia rosaria</i>	10	30

In every species observed, ten in all, a similar difference in the rate of pulsation exists, the rate in the sexually mature animal being in most cases about half that of the animal at the beginning or in very early stages of gonad development. By repeated stimulation the rate may often be somewhat increased, particularly in old animals, but the differences in animals under ordinary conditions are of the order of magnitude indicated. This dif-

ference in pulsation rate in itself indicates a very marked decrease in metabolic rate with advancing age. Mayer ('06) has noted that in the scyphomedusa *Cassiopeae* the rate of pulsation decreased with advancing age.

Young and old animals also differ in their behavior in another way. Under the usual conditions periods consisting of a larger or smaller number of pulsations alternate with periods of rest. The total length of pulsation periods as compared with rest periods decreases with advancing age; in other words the younger animal spends a larger portion of the time than the older in rhythmic pulsation. Pulsation periods may either be longer or more frequent or both in the younger animal than in the older. In *Aequorea* this difference is perhaps even more strongly marked than in other forms observed. In the large, sexually mature animals 60-80 mm. in diameter, when under natural and so far as possible constant conditions, the periods of quiescence are often one or two minutes long, sometimes even longer, the animal drifting passively during this time without a single pulsation. Following such a quiescent period a single pulsation or a pulsation period may occur. Under the same conditions the quiescent periods in young animals 15-20 mm. in diameter are usually very much shorter, commonly only a few seconds and the pulsation periods are usually longer than in the old animal. The young animal is then more continuously active and gives the impression of a much greater degree of "spontaneity," since it is usually impossible to distinguish any external exciting factor responsible for the beginning of pulsation after a period of quiescence. In the other species observed similar differences between young and old exist, but periods of quiescence are usually shorter than in *Aequorea*.

The irritability of the young animal as indicated by the effectiveness of direct mechanical stimulation in inducing pulsation during a quiescent period is distinctly greater than that of the old. Usually the quiescent animal in the earliest stages of gonad-development responds by pulsation to the slightest touch in the marginal region, while the sexually mature animals commonly respond only to much more intense mechanical action.

These differences between younger and older medusæ show of

course a considerable range of variation in different individuals, but are nevertheless striking features of the behavior of these animals when attention is directed to them and they remind one irresistibly of age differences in behavior in much higher animals. The apparent restlessness and spontaneity, the more rapid pulsation rate, and the greater irritability of the young animal as compared with the old all suggest that in these, as in other organisms, so far as known, the life history of the individual is from an early stage a process of physiological senescence which expresses itself dynamically as a decrease in rate or intensity of the fundamental energy-liberating metabolic reactions.

SUSCEPTIBILITY.

In determining the susceptibility of the animals various other criteria besides death and disintegration may be used as a check, viz., cessation of rhythmic pulsation, the disappearance of muscular contractility and shrinkage of jelly in some agents. As regards the relation between susceptibility and age all of these criteria give the same results. It is usually difficult or impossible to determine the exact time when an animal ceases to respond to stimulation by rhythmic pulsation or by slow muscular contractions and the early stages of shrinkage and of disintegration are not less difficult to determine. The times given in the following tables represent approximations only. Frequently pulsation or contractility is present at one observation and absent at the next, and the time of its disappearance can only be estimated. Nevertheless there is no difficulty in distinguishing the differences in susceptibility between young and old animals. The experiments were mostly performed with single pairs, one young, one old, but in some cases two or three of each were used.

Æquorea cærulescens.

This, being one of the most abundant species, was used to a large extent. The smallest and youngest individuals found were 8-10 mm. in diameter, without gonads, the largest, and oldest, sexually mature animals 60-75 mm. in diameter. The chief results are tabulated below. In these tables the sign \neq following a given time indicates that this is estimated from two observa-

tions between which the change in question occurred, and . . . ? indicates that the change or process did not begin until after the close of the experiment. In this species cessation of pulsation, disintegration and in some cases loss of contractility are available as criteria of susceptibility. The jelly is very firm and does not shrink to any great degree in any agent used.

The three tables agree in showing a greater susceptibility in young than in old animals, as indicated by cessation of rhythmic pulsation, loss of muscular contractility and disintegration. Special attention must be called to certain points. In Table II.

TABLE II.

Æquorea: SUSCEPTIBILITY TO KNC.

Series.	Diameter in Mm.	Conc. KNC.	Duration of Experiment.	No Pulsation after Stimulation.	No Contraction after Stimulation.	Disintegration.
I	A..	7	$m/10000$ After neutral red	1 hr.	1 min. \pm	5 min. \pm
	B..	12			5 " "	10 " \pm
	C..	20			10 " "	20 " \pm
	D..	50			40 " "	60 " \pm
2	A..	12	$m/10000$	4 hrs.	30 sec.	...?
	B..	30			20 min.	...?
I.	A..	18	$m/500$	17 hrs.	2-5 min.	12 hrs. \pm
	B..	60			10-15 "	17 " \pm
II.	A..	18	$m/200$	6 hrs.	3 min.	4-6 hrs.
	B..	60			8 "	6-? "
III.	A..	16	$m/100$	2 hrs.	At once	1 hr.
	B..	70			" "	2 "
IV.	A..	20	$m/100$	3 hrs.	At once	$1\frac{1}{4}-1\frac{3}{4}$ hrs.
	B..	60			" "	$1\frac{1}{4}-3$ "

it is of interest to note that KNC $m/10000$ is about as effective, or in some cases more effective than much higher concentrations, $m/500$, $m/200$, in stopping pulsation. It has been observed in other cases (Child, '16a, '16b) that rapidity of action of KNC and certain other agents, as indicated by death or other effects, increases only slightly with increase in concentration above a

certain limit or may even decrease. The facts suggest that the high concentrations may decrease permeability to themselves, at least up to a certain limit where their action becomes violently destructive to protoplasm, and so practically instantaneous.

In Table III., HCl and Table IV. KOH, the action in stopping pulsation increases very rapidly with increasing concentration,

TABLE III.

Æquorea: SUSCEPTIBILITY TO HCl.

Series.	Diameter in Mm.	Conc. HCl.	Duration of Experiment.	No Pulsation after Stimulation.	No Contraction after Stimulation.	Disintegration.
II.	18	$m/1000$	5 days	4 $\frac{1}{2}$ days	...?	...?
	60			...?	...?	...?
III.	15	$m/1000$	5 days	3 days —	4 days +	...?
	40			...?	...?	...?
3	18	$m/800$	5 days	3 days +	4 days \pm	...?
	60			...?	...?	...?
IV.	18	$m/500$	21 hrs.	At once		8-12 hrs.
	60			1 min. \pm		17 hrs. ...?
V.	18	$m/500$	1 hr.	At once		
	60			2 min.		
4	18	$m/400$	1 hr.	At once		
	60			" "		
6	18	$m/300$	2 $\frac{1}{4}$ hrs.	At once		55 min. ...?
	60			" "		...?
I.	18	$m/300$	2 $\frac{1}{4}$ hrs.	At once		55 min. ...?
	60			" "		2 $\frac{1}{4}$ hrs. ...?
5	18	$m/300$	2 $\frac{1}{4}$ hrs.	At once		
	60			1 min.		

proportionately much more rapidly than the concentration, because more or less acclimation to the lower concentration occurs. In HCl $m/1500$ —not tabulated—acclimation occurs in the young animal to a greater degree than in the old. After five days in this concentration the young animal still shows well-marked rhythmic pulsation after mechanical excitation while

the old animal cannot be induced to pulsate. In concentrations of $m/1000$ or higher, the young animal is always the more susceptible (see Table III.).

The records on the loss of muscular contractility are very incomplete, but it is evident that rhythmic pulsation and general

TABLE IV.
Æquorea: SUSCEPTIBILITY TO KOH.

Series.	Diameter in Mm.	Conc. KOH.	Duration of Experiment.	No Pulsation after Stimulation.	Disintegra- tion.
9 IV.	16	$m/1000$	84 hrs.	12 hrs. \pm	48 hrs. \pm
	60			48 hrs. \pm	...?
8 I.	18	$m/800$	34 hrs.	10 min.	4-6 hrs.
	60			24 hrs.	...?
8 IV.	15	$m/800$	28½ hrs.	5 min.	3-7 hrs.
	60			26 hrs. \pm	28½ hrs. ...?
9 III.	14	$m/600$	9 hrs.	2 min.	4-6 hrs.
	60			4-5 min.	9 hrs. ...?
9 II.	18	$m/400$	9 hrs.	30 sec.	2-4 hrs.
	75			1 min. +	3-5 hrs. +
9 I.	18	$m/300$	5 hrs.	5 sec. \pm	1-2 hrs.
	70			20 sec. \pm	3-5 hrs.
8 II.	14	$m/200$	5 hrs.	At once	2-4 hrs.
	60			10-20 sec.	3-5 hrs.
8 III.	16	$m/200$	3½ hrs.	At once	$1\frac{1}{4}-2\frac{1}{4}$ hrs.
	60			20-30 sec.	$2-3\frac{1}{2}$ hrs.

muscular contraction are not the same thing. Contractility of the muscles of the umbrella persists in all three agents long after ability to pulsate has disappeared, except in concentrations so high that death is practically instantaneous.

A complete but temporary inhibition of pulsation lasting from a few seconds to several hours occurs in almost all cases in HCl $m/600$. Above $m/600$ inhibition is usually permanent. At

the lower limit of concentration for temporary inhibition only the young member of a pair is inhibited; with somewhat higher concentration both are inhibited for a short time but the young animal usually resumes pulsation before the old if the differences in age are not too extreme. As the concentration increases, the length of the inhibition period increases more rapidly in the young than in the old, until about the upper limit permanent inhibition occurs at once in the young animal while some slight pulsation may reappear sooner or later in the old. In short the age differences in susceptibility appear here as in other features of the action of external agents.

Experiments on recovery after temporary exposure to an agent showed that the relation between recovery and age is the same as in other forms examined. The reappearance of the pulsation response to stimulation is the criterion of recovery used. Where the concentration is not too high, the exposure too long or the difference in age between the animals too great, the young will recover before the old on return to water, but with higher concentrations longer exposures and greater differences in age the old animal recovers earlier and usually more completely than the young. These relations between recovery, age, concentration and period of exposure are of course merely a special case of the age-susceptibility relation. To higher concentrations or longer times of exposure the young animal is more susceptible than the old, but to low concentrations or short times of exposure it is able to adjust itself more rapidly or to recover more rapidly afterward than the old (Child '15, '16a). The degree of difference between young and old animals also differs widely with different agents, because acclimation to some agents occurs much more rapidly than to others. The observations on recovery and its limits are as yet only fragmentary and further work is necessary before a complete statement can be made.

Some of the data of an experiment with HCl *m*/500 will serve as an example of recovery. Animals 15 mm. and 60 mm. are placed in HCl *m*/500 and at intervals a pair, one of each size, is returned to water. In the young animals all pulsation ceases at once in HCl, in the old it ceases after two minutes. On return to water after 5-10 minutes in HCl pulsation usually reappears

after 2-3 minutes in both old and young, often slightly earlier in the young. After one hour in HCl pulsation reappears after 10 minutes in the old, and after 12-13 minutes in the young and after 2½ hours in HCl pulsation reappears in the old after 25, in the young after 45 minutes. The longer the exposure the more the young animal falls behind the old in recovery. After 20 minutes in HCl $m/400$ young animals 18-20 mm. did not recover at all while old animals 60 mm. showed slight pulsation after four hours in water. Recovery after KNC is much slower and occurs only in much lower concentrations.

In all concentrations of KOH from $m/1000$ to about $m/250$ a primary acceleration of pulsation rate and increase in strength of contraction occurs, but in concentrations higher than $m/250$ pulsation usually ceases at once or in a few seconds. During this primary acceleration the pulsation rate is often doubled, but since the maximum rate usually persists only a few seconds and is followed by progressive decrease in rate it is difficult to determine whether the degree of primary acceleration differs in a characteristic way with age. It is certain, however, that the retardation following the acceleration occurs more rapidly in young than in old animals, so that pulsation ceases first in the young, as Table IV. indicates.

The opposite primary effects of acid and alkali on the pulsation rate are what might be expected in the light of what we know of their action on living protoplasm in general. In lethal concentrations, however, their action, following the primary effect, is essentially similar and the relations between susceptibility and age are the same for both.

Mitrocoma discoidea.

In this species the jelly is much less firm than in *Æquorea*, and marked shrinkage occurs in the young animals in acids and some other agents. Muscular contraction as distinguished from pulsation is much more frequent even in nature than in *Æquorea*. Frequently contraction brings about a folding of the umbrella with the two halves of the margin approximated or apposed along a straight line, or the margin may become square or very irregular in outline. The contracted condition may persist for several

minutes, or after very strong stimulation an hour or two before return to the usual form and resumption of pulsation occur. As in *Aequorea*, the ability to contract persists long after the ability to pulsate has disappeared.

In Table V. the chief data obtained with HCl are given. They are essentially similar to those for *Aequorea*.

TABLE V.
Mitrocoma: SUSCEPTIBILITY TO HCl.

Series.	Diameter in Mm.	Conc. HCl.	Duration of Experiment.	No Pulsation after Stimulation.	No Contraction after Stimulation.	Beginning of Shrinkage.	Disintegration.
X.	A 18	$m/1500$	114 hrs.	...?	Acclimated: active at end of expt.	...?	...?
	B 60			114 hrs. —			
II.	A 18	$m/1000$	55½ hrs.	40 hrs. ±	...?	...?	...?
	B 60			...?			
III.	A 18	$m/800$	48 hrs.	8½ hrs.	24 hrs. +	24 hrs.	40 hrs. ...?
	B 55			40 hrs. ±			
IV.	A 15	$m/700$	48 hrs.	At once	24 hrs. ±	30 hrs. ±	40 hrs. ...?
	B 60			15 hrs.			
5	A 20	$m/650$	34 hrs.	2 hrs.	24 hrs. ±	22 hrs.	30 hrs. ...?
	B 45			15 hrs. ±			
VI.	A 18	$m/650$	52½ hrs.	26 hrs. ±	24 hrs. ±	32 hrs.	45-52½ hrs.
	B 60			34 hrs. ±			
VII.	A 18	$m/600$	51½ hrs.	At once	24 hrs. ±	32 hrs.	45-51½ hrs.
	B 60			26 hrs. ±			
VIII.	A 18	$m/500$	4½ hrs.	At once	1 hr. ±	2 hrs.	...?
	B 60			" "			
IX.	A 20	$m/500$	9¾ hrs.	" "	At once	...?	...?
	B 60			" "			

The first series 5, X. in HCl $m/1500$ shows acclimation as regards the effect on pulsation rate, pulsation persisting longer in the young than in the old. In *Aequorea* also some degree of acclimation usually occurs in this concentration (p. 53). The

long persistence of pulsation in both members of the pair in Series 5, VI. (HCl $m/650$) as compared with the other series is anomalous and no reason for it is known. Otherwise the data in the table are consistent and show that for the concentrations included, except $m/1500$, the young animal is more susceptible than the old.

Results with KNC show similar relations between age and susceptibility, of course with much lower concentrations. The results of one series on pulsation susceptibility are given in Table VI.

TABLE VI.

Mitrocoma: SUSCEPTIBILITY OF PULSATION TO KNC.

Series.	Diameter in Mm.	Conc. KNC.	Duration of Experiment.	Pulsation.
4. IV.	18	$m/100000$	5 hrs.	Pulsation only retarded: rate $A = \frac{1}{2}$ rate B .
	60			
4. III.	18	$m/50000$	$5\frac{1}{2}$ hrs.	Very slight, irregular.
	60			Well-marked, regular.
4. I.	16	$m/25000$	$\frac{1}{2}$ hr.	Ceases in 10-15 min.
	60			" " 15-20 "
4. V.	18	$m/10000$	$\frac{1}{2}$ hr.	Ceases in 5 min. ±
	55			" " 15 " ±

In the first pair, 4, IV., $m/100000$, the susceptibility is shown, not in the time of cessation of pulsation, but in the retardation of the rate. At the beginning of the experiment rate $A = 2$, rate B approximately, but after five hours rate $A = \frac{1}{2}$ rate B . In the second pair, 4, III., $m/50000$, pulsation in the young animal is almost completely inhibited after $5\frac{1}{2}$ hours, while in the old it is still strong. Here rate A was very much lower than rate B . In the higher concentrations pulsation ceases in a few minutes.

A few experiments with other agents give similar results. In neutral red animals 18-20 mm. long cease to pulsate, undergo shrinkage and begin to disintegrate much earlier than animals 60 mm., although no marked difference in rate or intensity of staining is apparent. In methylene blue $1/30000$, increased to

1/15000 after 24 hours, the animals live for several days with all cellular tissues stained. After 48 hours pulsation rate $A = \frac{1}{2}$ pulsation rate B approximately, where A and B are animals of 18–20 and 50–60 mm. respectively.

Phialidium gregarium.

The youngest animals found were mostly 7–8 mm. in diameter, while the old, sexually mature individuals measured 20–25 mm. Only a few series of susceptibility experiments were made with this species, HCl $m/1500$, $m/1000$, $m/800$, $m/600$, neutral red and methylene blue being used. The susceptibility relations are similar to those observed in the other species. In this rather delicate form shrinkage is a characteristic feature of the action of various agents, the younger animals undergoing shrinkage earlier and to a greater degree than the old, a decrease of half or three fourths in diameter occurring in many cases.

Sarsia rosaria.

In this species the umbrella is very deep, the oral-aboral axis being almost or quite twice the diameter. Depth is therefore a more convenient measure of size than diameter and is commonly used. The sexually mature animals average about 15 mm. in depth of umbrella, but the extremes range from 10–12 mm. on the one hand to 20 mm. on the other. In the smallest and youngest animals found, 6–7 mm. in depth, the gonads had already begun to develop, and although the susceptibility of these forms is characteristically greater than that of the mature individuals the differences in age and in susceptibility in the animals available are not as great as in the other species examined.

In general, results with this species are similar to those with others but in some pairs an individual 12–15 mm. showed a susceptibility equal to or even greater than that of 7–8 mm. animals. Since none of the animals within these limits of size represent very great differences in physiological age, all being near sexual maturity, it is to be expected that occasionally a larger animal will show a higher susceptibility than a smaller.

The agents used were KNC $m/50000$, $m/10000$, $m/5000$, HCl $m/1000$, $m/600$, $m/500$, $m/400$, neutral red and methylene blue,

and the age differences appeared in all with the few exceptions above noted. One of these exceptions was rather remarkable. In KNC $m/50000$ animals 7-8 mm. usually lose the ability to pulsate, even on stimulation, in 3-10 minutes, animals 15 mm. after 1-2 hours, while in $m/10000$ the times for cessation of pulsation are respectively 1-2 minutes and 3-10 minutes. In one case, however, in KNC $m/10000$ an animal 15 mm. ceased to pulsate in three minutes, while an animal 8 mm. in the same closed container continued normal pulsation during thirty minutes, longer observation being impossible. For some reason this particular animal was practically insusceptible to KNC $m/10000$, a concentration which stops pulsation in the younger individuals of the other species examined in 5 minutes or less and in other younger individuals of *Sarsia* in 1-3 minutes. This extreme exception is merely recorded without any attempt to account for it. The observations on *Sarsia* do indicate a wider range of physiological condition in animals of a given size than in the other species examined, but they afford no clue to interpretation of a condition so extremely exceptional as this.

CONCLUSION.

The preceding data make it evident that the hydromedusa, like other animals, undergoes a progressive change in physiological condition with advancing development, the differences in behavior and susceptibility being indicators of this change. It has been shown that in other forms a decrease in the rate of oxidations is a characteristic feature of the change and the facts indicate that a similar decrease occurs here, although these forms are not very favorable material for the direct determination of oxygen consumption or CO_2 -production. The assumption that the differences in susceptibility are due merely to differences in permeability of surface membranes is refuted by the results obtained with neutral red and methylene blue and by the cases of acclimation. Moreover, there is every reason to believe that the age differences in susceptibility to cyanides are at least more directly associated with the oxidation rate than with permeability.

As regards permeability, however, it is becoming more and more evident to investigators in this field that the condition of

the membrane is not independent of the metabolic condition of the protoplasm. Permeability to various agents is itself within certain limits an indicator of age or physiological condition of the cell.

The progressive change in behavior, the decrease in pulsation-rate and "spontaneity" indicate very clearly a process of physiological senescence and the data on susceptibility only confirm and extend the facts of observation.

SUMMARY.

Four species of hydromedusæ show evidence of a process of physiological senescence in the decrease in pulsation-rate and apparent spontaneity and in the decrease in susceptibility to KNC, HCl, KOH and "vital" dyes in various concentrations with advancing development. The criteria of susceptibility are change in rate or cessation of pulsation, loss of contractility, shrinkage of the jelly and disintegration of cellular tissues, and all these criteria agree. The facts justify the conclusion that a decrease in the rate of metabolism, or more specifically of oxidations, is a characteristic feature of this process of senescence.

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